REMARKS

Claims 32-43 are pending.

Claims 32-43 were rejected under 35 USC 103(a) for purportedly being unpatentable over Malek et al (U.S. Pat. No. 5,130,238) in view of Kenten et al (U.S. Pat. No. 6,174,709).

Applicants respectfully traverse. Malek et al. do not teach or suggest the presently claimed subject matter. Malek et al. merely teaches a few variations of the NASBA protocol. Nothing in the disclosure of Malek teaches or suggests that an ECL probe can be used in NASBA amplification. Neither does Malek teach or suggest using a capture probe labeled with binding species and bead coated with binding species complementary to the capture probe. Also Malek does not teach or suggest providing conditions of temperature and buffer to allow the hybridization of the probe and an RNA template and the binding of the binding species on the capture probe with the complementary binding species on the bead to form a bead bound complex and detecting the bead bound complex using ECL detection. Moreover, Malek does not teach or suggest the desirability of using ECL labels and ECL detection. All of these deficiencies of Malek are admitted by the Examiner (Office Action, page 6).

Applicants submit that Kenten does not compensate for the deficiencies of Malek. More specifically, Kenten does not teach or suggest the use of ECL probes in the NASBA amplification of Malek. Applicants urge that the claimed embodiment of the invention (the detection of unlabeled amplification products through the use of two probes, one having an ECL label and the other having a capture moiety claimed in Claims 32 and 38 steps (c-e) is not disclosed in the Kenten reference. By contrast, Kenten et al disclose the combination of a labeled primer and a single labeled probe.

Furthermore the Examiner's statement that "In response to applicant's argument that the references fail to show certain features of applicant's invention, it is noted that the features upon which applicant relies (i.e., (a) the invention does not require sample pretreatment and (b)the detection of unlabeled amplification products through the use of two probes, one having ECL label and the other having a capture moiety) are not recited in the claims" (Office Action, page 9) is incorrect.

For example, the detection of unlabeled amplification products through the use of two probes, one having ECL label and the other having a capture moiety is specifically recited in claims 32 and 38 steps c-e. (See, Claim 32 and 38).

In addition, claims 32 and 38 form second mixtures by adding the probes to the amplification reaction mixture without the removal of the amplification enzymes prior to the addition of the probes. The fact that the amplification mixture does not require pretreatment before the probes are added is surprising and unexpected because, at a minimum, the probe sequence could be modified by the very enzymes and components required in the amplification medium. These components are not present in the amplification of Kenten. According to the present invention three separate enzymes (i.e., reverse transcriptase, RNase H, and an RNA polymerase) are present simultaneously in the assay, which creates a greater likelihood of unwanted side reactions taking place without pretreatment of the sample to remove contaminants. By contrast, PCR assays use a much simpler mix of enzymes that would be less likely to interfere with the reaction and require, at a minimum, denaturation of the double stranded product. A person of ordinary skill in the art would expect hybridization efficiencies to RNA in an isothermal NASBA system to be quite low, especially when two hybridizations are required, because of the lack of denaturation step and the complex structure and folding of RNA

molecules known in the art. The intramolecular hybridization within the RNA strand makes hybridization to external probes more difficult. In addition, the lower stability of RNA relative to DNA might have been expected to negatively interfere with the detection of the amplification product.

Applicants respectfully submits that there is no teaching in Kenton where a person of ordinary skill in the art could find suggestion or motivation to use the detection method of Kenten with the NASBA amplification. If the Examiner disagrees, the Examiner is respectfully requested to specifically point out where such suggestion or motiviation is. The citation relied on by the Examiner was taken out of context and does not teach or suggest the desirability of making the combination suggested.

The Examiner relies on the following statement: "The unexpected exponential amplification of the invention greatly simplifies the process of amplifying multiple nucleic acid sequences of interest present in a sample (Column 5, lines 1-4)" (Office Action, page 8). Applicants respectfully submit that this reliance is misplaced.

Kenten teaches a single primer amplification, where the amplification is expected to be linear and not exponential. In reviewing prior art, Kenten states:

> [...] if a single unpaired primer is used in place of two (paired) primers, the result is a linear growth in extension product copy number instead of an exponential growth of both strands (3). It is generally believed that the reason for the linear growth in copy number with cycle using a single unpaired primer is that only the template strand is replicated during each cycle. The primer extension itself is not copied.

[Kenten, col. 1, lines 54-61].

Kenten specifically teaches that an exponential amplification using a single unpaired primer is the advantageous discovery of his invention:

> [...] heretofore unavailable method for achieving exponential amplification of a specific nucleic acid sequence of interest requiring only a single primer but retaining specificity of action would be an important and unexpected contribution to the art.

[Kenten, col. 4, lines 8-12].

The unexpected exponential character of the single primer PCR amplification in Kenten can not be viewed as a suggestion or motivation to combine probes of Kenten with NASBA amplification of Malek. Kenten does not teach or suggest that a single primer PCR amplification is in any way related to or can be a substitute for NASBA, or that such a substitution may be desirable. Although Kenten discloses the use of ECL probes in PCR-based single primer amplification, the differences between NASBA and PCR reactions are substantial and the skilled artisan would not reasonably expect that the successful incorporation of ECL technology with one would be a good predictor of success with the other.

Thus, Applicants urge that there is no suggestion or motivation to combine the teachings of Malek et al with the teachings of Kenten et al. Hence, a prima facie case of obviousness has not been established. More specifically, there is no factual support for suggestion or motivation for using a detection method of Kenten with the NASBA amplification of Malek. The suggestion to combine the elements must come from the reference cited and not from the applicant's disclosure. See *In re Dow Chemical Co.*, 837 F.2d 469, 473, 5 USPQ2d 1529, 1531 (Fed. Cir. 1988). To establish a prima facie obviousness based on a combination of references, the Examiner is required to demonstrate that the prior art provide "a reason, suggestion, or motivation to lead an inventor to combine those references." *Pro-Mold and Tool Co. v. Great Lakes Plastics Inc.*, 75 F.3d 1568, 1573, 37 USPQ2d 1626, 1629 (Fed. Cir. 1996).

> [E]vidence of a suggestion, teaching, or motivation to combine may flow from the prior art references themselves, the knowledge of one of ordinary skill in the art, or in some cases, from nature of the problem solved. ... The range of sources available, however,

does not diminish the requirements for actual evidence. That is, the showing must be clear and particular.

In re Dembiczak, 175 F.3d 994, 999, 50 USPQ2d 1614, 1617 (Fed. Cir. 1999) (Citation omitted, emphasis added).

Applicants respectfully maintain that Malek and Kenten, either taken alone or in combination, do not provide guidance or motivation for the successful use of ECL technology with NASBA technology. Therefore, withdrawal of this rejection under 35 USC 103(a) is respectfully requested.

If there are any further points requiring attention prior to allowance or if these remarks do not place the rejected claims in condition for allowance, then Applicants respectfully request the courtesy of a telephonic interview.

No additional fee is required. If there any such fees, please charge them to our firm Deposit Account No. 50-0540.

Respectfully submitted,

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